

liver be examined as a potential source. This speculation is interesting in the light of earlier comments by Ashman *et al.* (1978) on this particular problem when we stated "Although it is possible that another organ may take over the functions of the thymus in neonatally-thymectomized quokkas, it is difficult to see why this should function only early in life. Perhaps the thymus acts primarily as a biological amplification system, by providing a favourable environment for the proliferation of precursors migrating in from another source. In the absence of the thymus, these precursors may undergo spontaneous differentiation into clones of immunocompetent cells."

References

- Ashman, R. B., Keast, D. and Waring, H. (1971).—The evaluation of the phytohaemagglutinin (PHA) response of peripheral leucocytes and skin graft rejection in thymectomized quokkas (*Setonix brachyurus*). *Proc. Aust. Soc. Med. Res.*, **2**: 454.
- Ashman, R. B. and Papadimitriou, J. M. (1975h).—Development of lymphoid tissue in a marsupial, *Setonix brachyurus* (quokka). *Acta Anat.*, **91**: 594-611.
- Ashman, R. B., Stanley, N. F. and Waring, H. (1978).—Immunobiology of marsupials. In: *Animal Models of Comparative and Developmental Aspect of Immunity and Disease*, M. E. Gershwin and E. L. Cooper (eds.), 154-165, Pergamon Press, Oxford.
- Bell, R. G., Lynch, N. R. and Turner, K. J. (1974).—Immunoglobulins of the marsupial *Setonix brachyurus* (quokka): characterization of three major serum classes immunoglobulin G-2, immunoglobulin G-1 and immunoglobulin M. *Immunology*, **27**: 1103-1115.
- Block, M. (1964).—The blood forming tissues and blood of the newhorn opossum (*Didelphys virginiana*). *Ergebnisse der Anatomie u. Entwicklungsgeschichte*, **37**: 1-237.
- Charlick, J., Manassis, C., Stanley, N. F., Waring, H. and Cockson, A. (1981).—Quantitative alterations of the aerobic bacterial flora of the pouch of *Setonix brachyurus* during oestrus, anoestrus, pregnancy and lactating anoestrus. *Aust. J. exp. Biol. med. Sci.*, **59**: 743-751.
- Cockson, A. and McNeice, R. (1980).—Survival in the pouch: the role of macrophages and maternal milk cells. *Comp. Biochem. Physiol.*, **66A**: 221-225.
- Jurgelski, W., Hudson, P. M. and Falk, H. L. (1976).—Embryonal neoplasm in the opossum: a new model for solid tumours of infancy and childhood. *Sci.*, **193**: 328-332.
- Lightowlers, M. W. (1979).—Thymus function in the quokka: an analysis of mammalian thymus-dependent immunity. Ph.D. Thesis, Department of Zoology, University of W.A.
- Lynch, N. R. and Turner, K. J. (1974a).—Time course of homocytotropic and passive haemagglutinating antibody synthesis in the marsupial *Setonix brachyurus*, the quokka. *Int. Archs Allergy appl. Immun.*, **47**: 933-941.
- Lynch, N. R. and Turner, K. J. (1974b).—Immediate hypersensitivity responses in the marsupial *Setonix brachyurus* (the quokka). Biological properties of the homocytotropic antibody. *Aust. J. exp. Biol. med. Sci.*, **52**: 425-432.
- Lynch, N. R. and Turner, K. J. (1974c).—Immediate hypersensitivity responses in the marsupial *Setonix brachyurus* (the quokka). Characterization of the homocytotropic antibody. *Aust. J. exp. Biol. med. Sci.*, **52**: 755-766.
- Miller, J. F. A. P. (1962).—Immunological significance of the thymus of the adult mouse. *Nature*, **195**: 1318-1319.
- Stanley, N. F., Waring, H., and Yadav, M. (1966).—The thymus: experimental and clinical studies. In: *Ciba foundation Symposium*, G. E. W. Wolstenholme and R. Porter (eds.), 207-213. Churchill, London.
- Stanley, N. F., Yadav, M., Waring, H. and Eadie, M. (1972).—The effect of thymectomy on response to various antigens of a marsupial, *Setonix brachyurus* (quokka). *Aust. J. exp. Biol. med. Sci.*, **50**: 689-702.
- Yadav, M. (1969).—The thymus glands of the (quokka) *Setonix brachyurus*, and their role in immunological processes. Ph.D. Thesis, Department of Zoology, University of W.A.
- Yadav, M. (1971).—The transmission of antihodies across the gut of pouch young marsupials. *Immunology*, **21**: 839-851.
- Yadav, M. (1973).—The presence of the cervical and thoracic thymus lobes in marsupials. *Aust. J. Zool.*, **21**: 285-301.
- Yadav, M. and Eadie, M. E. (1973).—Passage of maternal immunoglobulins to the pouch young of a marsupial, *Setonix brachyurus*. *Aust. J. Zool.*, **21**: 171-181.
- Yadav, M. and Papadimitriou, J. M. (1969).—The ultrastructure of the neonatal thymus of a marsupial. *Setonix brachyurus*. *Aust. J. exp. Biol. med. Sci.*, **47**: 653-668.
- Yadav, M., Stanley, N. F. and Waring, H. (1972a).—The thymus glands of a marsupial, *Setonix brachyurus* (quokka), and their role in immune responses. Structure and growth of the thymus glands. *Aust. J. exp. Biol. med. Sci.*, **50**: 347-356.
- Yadav, M., Stanley, N. F. and Waring, H. (1972h).—The thymus glands of a marsupial, *Setonix brachyurus* (quokka), and their role in immune responses. Effects of thymectomy on somatic growth and blood leukocytes. *Aust. J. exp. Biol. med. Sci.*, **50**: 357-364.

Microbiology studies on Rottneest Island

by N. F. Stanley and J. S. Mackenzie

Department of Microbiology, University of Western Australia

Abstract

Studies of four different groups of viruses, the bacterial flora of the pouch, and the ecology and pathogenesis of quokka filariasis, comprise the items of this review. The presence of reoviruses and arboviruses confirm the ubiquity of two major virus groups spread by entirely different means. The epidermal papillomata of the tails and feet of quokkas have been shown to be associated with a 'new' poxvirus. The Gram-negative bacterial flora of the pouch was dramatically reduced prior to birth. The life-cycle and immunobiology of quokka filariasis have been defined for the first time. Influenza A virus studies of birds have led to the isolation of a virus with a novel antigenic configuration.

Introduction

Microbiological (virology, bacteriology, parasitology, serology) studies on Rottneest Island were initiated for several reasons, the main one being the immunological immaturity of the quokka pouch-young in the first few days of pouch life. The relative isolation of the island quokkas for several thousand years, their constant association with seabirds and arthropods, and their more recent association with man, has led to the studies briefly referred to here.

This review comprises studies of the following:

- A. Epidemiology of reovirus infections, arbovirus infections and avian influenza.
- B. Quokkapox—a 'new' poxvirus.
- C. The bacterial flora of the marsupial pouch.
- D. Filariasis in the quokka.

A.1 Epidemiology of reovirus infections

Reoviruses are double-stranded RNA viruses of great ubiquity but little potential to produce diseases in man or animal (Stanley 1974, 1977, 1981). Antibodies to the three antigenic types have been found in all terrestrial vertebrates tested (Stanley *et al.* 1964, Stanley 1967). Studies on the Rottneest Island quokka have shown that quokkas in continuous contact with man manifest higher levels of antibody conversion than those with little human contact (Stanley and Leak 1963) (see Table 1). These observations led us to suggest that reoviruses infecting man could be transmitted to animals in contact with man; since that observation we have obtained evidence which does not support this hypothesis, by isolating reoviruses and demonstrating antibodies to them on Bald Island quokkas.

A.2 Epidemiology of arbovirus infections

Approximately 450 arboviruses have been described, of which about one-quarter produce disease in man and animals. All have the ability to replicate in both the arthropod vector and the vertebrate host reservoir and the majority are spread by mosquitoes, ticks and culicoides. They include serious pathogens of man, such as Australian encephalitis, yellow fever and dengue, and of animals, such as bluetongue and bovine ephemeral fever. One way of detecting the presence of these viruses is by searching for antibodies to them in the serum of vertebrates.

Table 1

Percentages of quokkas with detectable H-1 antibody (>1/10) against reovirus types 1, 2 and 3

Locality	No. of animals	Percentage positive against reovirus type			Contact with man
		1	2	3	
West End, Rottneest Island	35	17	34	28	rare
Lakes-Settlement, Rottneest Island	27	67	89	93	frequent
Mainland	10	70	100	100	continuous

After Stanley and Leak (1963)

In our studies of Australian encephalitis (Stanley 1975, 1980), we examined sera from many vertebrates throughout the State, including the Rottneest Island quokka. Sixty-five per cent of 87 sera tested in 1975 showed the presence of antibodies to flaviviruses—a group of arboviruses containing the pathogens Murray Valley encephalitis and Japanese B encephalitis. The identity of the virus has not been determined. Similar tests on Garden Island tammar were negative.

A.3 Epidemiology of avian influenza

As part of the WHO's International Expanded Programme on the Ecology of Influenza A viruses, over 6 000 wild birds have been examined for the presence of influenza throughout Western Australia, including 234 birds of Rottneest Island (Mackenzie, Edwards, Holmes and Britten, unpublished results). Of these, 166 cloacal swabs were obtained from *Puffinus pacificus* (wedge-tailed shearwater), 10 swabs from *Larus novaehollandiae* (silver gull), 15 swabs from *Sterna bergii* (crested tern), and 43 swabs from *Tadorna tadornoides* (Australian shelduck). Three strains of avian influenza have been isolated, all from the shelduck, and have been subtyped by Dr Hinshaw, International Reference Laboratory for Animal Influenza, Memphis, U.S.A., as H? N2, Hswl Nav6 and H? Nav6. The latter two viruses are of particular interest; the Hswl Nav6 isolate represents a novel antigenic combination which has not been found previously anywhere in the world; and the H? Nav6 isolate appears to have a haemagglutinin which does not react with any reference antisera, and may represent a new, as yet undescribed, haemagglutinin. If this is indeed the case, the possible surface antigens of influenza A will be extended to 14 haemagglutinin types and 9 neuraminidase subtypes (Hinshaw, Webster and Rodriguez, 1979). The third isolate, H? N2, has not yet been fully subtyped. No isolates of Newcastle disease virus were found.

B. Quokkapox—a "new" poxvirus

A pox-like virus has been implicated as the aetiological agent of epidermal papillomata commonly found on the tails of quokkas, and more rarely on other extremities such as nose, ears and feet. The papillomata usually occur on the dorsum of the distal end of the tail as single, but sometimes multiple, wart-like lesions varying in size from a few mm to 5-6 cm. Electron microscopic examination revealed virus particles morphologically resembling mature and immature poxviruses in the cytoplasm of cells of the stratum granulosum (Papadimitriou and Ashman 1972).

Extensive attempts have recently been made to cultivate the virus from homogenates of papillomata (Lalor, Mackenzie and Stanley, unpublished results), including inoculation onto chorioallantoic membrane of 8-, 10-, 12- and 14-day embryonated chicken eggs, into brains of suckling mice, and in tissue cultures of avian (chick embryo fibroblasts), eutherian (BHK-21 cells, vero cells and mouse L-cells) and marsupial (quokka kidney, lung, skin and brain; *Sminthopsis crassicaudata* and *Wallabia bicolor* pouch-young cell lines) origin, but no evidence of virus replication or cytopathic effect was observed in any culture over four consecutive passages.

Co-cultivation experiments were also unsuccessful. Nevertheless, quokkapox virus in papillomata homogenates was able to reactivate heat-inactivated vaccinia virus on the chorioallantoic membrane and in vero cells, which would indicate that the virus was able to enter cells and replicate to a limited extent. The property of reactivation is characteristic of poxviruses, regardless of their subgrouping (Joklik 1966), and thus confirms the morphological identification by electron microscopy that the virus belongs to the poxvirus family.

Attempts to subgroup quokkapox serologically within the poxvirus family have been hindered by the lack of a purified antigen and of a specific, high titre, antiserum. However, in preliminary experiments using a crude antigen preparation from papillomata homogenates, a slight cross-reaction was observed between quokkapox and fowlpox in a complement-fixation test, but not between quokkapox and vaccinia. The level of cross-reaction, which was similar to that found between fowlpox and Western Australian wild bird poxviruses (Annuar and Mackenzie, unpublished results), suggests that quokkapox may be a member of the avipox subgroup, but further experiments are required to substantiate this classification.

C. The bacterial flora of the marsupial pouch

The bacterial flora of the gut of the pouch-young and the pouch of the quokka were initially studied by Yadav *et al.* (1972) in an endeavour to determine the nature of the microbial environment presented to the immunodeficient embryo at birth. The bacterial flora was abundant and complex in all animals tested except a pregnant mother just prior to birth. This observation posed some interesting questions of possible hormonal control of bacteria in the pouch. Further and more exacting studies by Charlick *et al.* (1981) defined quantitative and qualitative differences in the pouch flora in different stages of the oestrous cycle. Oestrus was characterized by high numbers of Gram-negative bacilli, and during non-lactating anoestrus, a predominantly Gram-positive flora was found. A marked selective decrease in Gram-negative flora occurred during the gestation period until they were virtually non-existent in the pouch prior to birth. This interesting and dramatic phenomenon awaits explanation.

D. Filariasis in the quokka

Breinlia macropi, the peritoneal filaria of the quokka, has hitherto been known largely by its adult morphology: nothing of its life-cycle or immunobiology in its host has been established. Scanning electron microscopy (SEM) revealed certain features of adult *B. macropi* morphology which previously were not discernible under light microscopy. The cuticular ornamentation, for instance, differs from that of a closely-related species, thus auguring a diagnostic potential among *Breinlia* species. Similarly, SEM has detailed the precise, diagnostic topography of the caudal extremity of the infective larva.

Our studies (Yen 1982) have shown that *Aedes camptorhynchus* is the natural vector of *B. macropi*, the larvae of which develop within the fat-body of this mosquito. Although other mosquito species

are susceptible to the parasite's infection, only *Ae. camptorhynchus*, with a natural infection rate peaking at 22%, is capable of transmitting *B. macropi* infection among the quokkas on Rottnest Island. Transmission of the parasite on the island is limited to between June to October, coinciding with the transient availability of brackish water for *Ae. camptorhynchus* to breed. Despite this, 95 of 203 (46.8%) of quokkas from the island were microfilaraemic, with males having significantly higher microfilarial rate than females. Although this difference in infection rate between the sexes was not apparent by fluorescent antibody (IgG) detection, this serological method detected 93% infection in the 203 animals examined. Both the parasitological and the serological data correlate well with the detection of circulating antigens as determined by the ELISA and counter-immunoelectrophoresis methods.

Peripheral blood leucocytes from normal, microfilaraemic and amicrofilaraemic quokkas proliferated equally well in dose-dependent pattern to PHA and CON A *in vitro*. No amount of soluble microfilarial, adult worm or secretory-excretory antigens, however, could elicit a significant response from these leucocytes. This was despite exhaustive serum or cell treatment.

By contrast, peritoneal macrophages from normal, amicrofilaraemic or microfilaraemic quokkas were equally efficient in adherence and cytotoxicity to *B. macropi* microfilariae *in vitro* in the presence of infected (microfilaraemic and amicrofilaraemic) sera. The prerequisite heat-labile serum factor probably resides in the reaginic antibody of the quokka. This factor, present only in the infected sera, seems to be cytophilic and specifically sensitized the macrophages to initiate adherence and cytotoxicity. As apparent from SEM, TEM, and time-lapse video recording, the death of the microfilariae is preceded by macrophages flattening themselves onto the microfilariae, encircling them and secreting material onto their surface. This killing process is effected by at least two types of macrophages distinguishable by size and ruffling of their membranes. The killing is also accompanied by inter-digitation of macrophage membranes of adjacent cells which jointly encircle the microfilariae as a cylinder of cells. The presence of multinucleate cells has also been noted in this study.

Conclusion

The microbiological studies cited show that the Rottnest Island quokka is not microbiologically unique, but that selected studies of an island-marsupial ecology can provide meaningful data which add significantly to our understanding of host-parasite relations. This is seen with the remarkable reduction in the bacterial flora of the pouch prior to birth, the clear demonstration of the cycle of filaria, *Ae. camptorhynchus* and the quokka, and the demonstration of a 'new' poxvirus.

References

- Charlick, J., Manassis, C., Stanley, N. F., Waring, H. and Cockson, A. (1981).—Quantitative alterations of the aerobic bacterial flora of the pouch of *Setonix brachyurus* during oestrus, anoestrus, pregnancy & lactating anoestrus. *Aust. J. exp. Biol. Med.*, 59: 743-757.
- Hinshaw, V. S., Webster, R. G. and Rodriguez, R. J. (1979).—Influenza A viruses: combinations of hemag-

- glutinin and neuraminidase subtypes isolated from animals and other sources. *Arch. Virol.*, **62**: 281-290.
- Joklik, W. K. (1966).—The poxviruses. *Bact. Rev.*, **30**: 33-66.
- Papadimitriou, J. M. and Ashman, R. B. (1972).—A poxvirus in a marsupial papilloma. *J. gen. Virol.*, **16**: 87-89.
- Stanley, N. F. (1967).—Reoviruses. *Brit. med. Bull.*, **23**: 150-155.
- Stanley, N. F. (1974).—The reovirus murine models. *Progr. med. Virol.*, **18**: 257-272.
- Stanley, N. F. (1975).—The Ord River Dam of tropical Australia. In: *Man-made Lakes and Human Health*. N. F. Stanley and M. P. Alpers (eds.), 103-112. Academic Press, London.
- Stanley, N. F. (1977).—Diagnosis of reovirus infections: comparative aspects. In: *Comparative Diagnosis of Viral Diseases, Vol. 1*. E. Kurstak and C. Kurstak (eds.), 385-421. Academic Press, New York.
- Stanley, N. F. (1980).—Man's role in changing patterns of arbovirus infections. In: *Changing Disease Patterns and Human Behaviour*. N. F. Stanley and R. A. Joske (eds.), 151-173. Academic Press, London.
- Stanley, N. F. (1981).—Reoviridae: orbivirus and reovirus infections of mammals and birds. In: *Comparative Diagnosis of Viral Diseases, Vol. IV*. E. Kurstak and C. Kurstak (eds.), 67-104. Academic Press, New York.
- Stanley, N. F. and Leak, P. J. (1963).—The serologic epidemiology of reovirus infection with special reference to the Rottnest Island quokka (*Setonix brachyurus*). *Amer. J. Hyg.*, **78**: 82-88.
- Stanley, N. F., Leak, P. J., Grieve, G. M. and Perret, D. (1964).—The ecology and epidemiology of reovirus. *Aust. J. exp. Biol. med. Sci.*, **42**: 373-384.
- Yadav, M., Stanley, N. F. and Waring, H. (1972).—The microbiological flora of the gut of the pouch young and the pouch of a marsupial, *Setonix brachyurus*. *J. Gen. Microbiol.*, **70**: 437-442.
- Yen, P. (1982).—Filariasis in the Australian marsupial, *Setonix brachyurus* (quokka). Ph.D. Thesis, Microbiology Dept., University of Western Australia.

The Rottnest experience

by G. Seddon

Faculty of Architecture and Planning, University of Melbourne

Abstract

This paper attempts to elucidate some of the diverse meanings that Rottnest has had for Western Australians. The psychological function of myth has always been to tell stories that establish exemplary models of behaviour and give dramatic substance to values, aspirations and conventions. Behaviour on Rottnest has many quasi-ritualistic elements, and the stories people tell about it are full of mythic overtones. Rottnest has played a significant part in sustaining Western Australians' self-image as a society that is friendly, gregarious, simple, unpretentious, physically oriented, pleasure loving and egalitarian. The paper explores some of these myths and their dependence on the physical characteristics of a particular place—the island of Rottnest.

Chronicle and myth

The history of Rottnest can be told in two ways. One is to give a chronicle of events. That has been done often for Rottnest. The broad outlines are very well known—the accounts of the early voyages of discovery, the settlement of Swan River Colony in 1829 and the plan for the township of Kingstown on the island in 1831 (from which the name of the Army settlement, Kingstown Barracks, is derived); the arrival of Constable Welch to take charge of Aboriginal prisoners in 1838 and the long subsequent history as a prison. An alternative use was foreshadowed by the increasing popularity of the island as a summer resort with successive Governors of the colony. Governor Fitzgerald (1848-1855) commandeered the original superintendent's cottage. Kennedy authorised the building of a new Government House on Rottnest and:

'a design was prepared by the Royal Engineers' Office in Perth and drawings were signed by Richard Roach Jewell, the Clerk of Public Works' (Commission for the National Estate, 1977, Book 1, p. 29).

The building was completed in 1864—although not precisely to these drawings—and first occupied by Governor Hampton.

Thus began the recreational use of Rottnest—at first for viceregal shooting parties, but in 1903 the prison was closed, the pilot station transferred to Fremantle, and the island proclaimed for public use in March 1907 by his Excellency Admiral Sir Frederick Bedford. The present and future primary use was then established. In the last half century a secondary use of the island has evolved as a laboratory for scientific research. This is not always compatible with unrestricted large scale recreational pressures, although of course science has a special rôle in monitoring the effects of such pressures.

The second and more difficult approach to the history of Rottnest is to attempt to elucidate its meaning. It undoubtedly has meaning. The island seems to hold a very special place in the minds and imaginations of Western Australians. It is treasured with a fierce affection that calls for explanation, which must form a significant element in any serious attempt at a cultural history of Western Australia. The complex of attitudes towards Rottnest appears to encapsulate many deep-lying aspirations about the good life, the nature of society, the proper relations of man with man and of man with the environment,